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ON THE BIOLOGICAL ASPECTS OF PAPER MILL POLLUTION IN CERTAIN PARTS OF THE COLUMBIA RIVER WATERSHED, WITH ESPECIAL REFERENCE TO 'SPHAEROTILUS NATANS'

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A Report

On the Biological Aspects of Paper Mill Pollution in Certain Parts of the Columbia River Watershed, with Especial Reference to Sphaerotilus Natans.

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1941

In 1940 some questions were asked the writer concerning the growth in the Columbia River near Portland, Oregon, of a "slime" growth which was causing gross clogging of the nets of commercial fishermen. Subsequent investigation by workers of the Health Departments of Washington and Oregon indicated the organism might be Sphaerotilus. On August 11, 1941 the writer was detailed by the U. S. Public Health Service, at the request of the Health Officials of these two states, to make a field investigation of the organism and the problems involved.

It may be recorded that Sphaerotilus or similar bacteria have given a great amount of trouble in streams, especially in the Scandinavian countries where the paper pulp industries are important; also in streams where industrial wastes of various types are pollutants; and in sewage disposal plants of the activated sludge type. Sphaerotilus and similar bacteria or fungi grow in huge slimy masses, attached to any submerged object, and grow in both heavily and lightly polluted waters. Reproduction is abundant and growth is very fast. Most of the living portion of older flocs is peripheral, but the basal or distal portion of the floc is resistant to disintegration, and forms a matrix and probably a substrate for the entanglement and/or growth of a multiplicity of other debris and organisms, until a disproportionate size is attained.

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Sphaerotilus itself will grow with little dissolved oxygen, although its maximum development is in the presence of an ample supply. Apparently its preferred substrate for heaviest production consists of poly- and di-saccharoses, although other carbonaceous materials are utilized. For nitrogen, there is some indication that amino acids are necessary although nitrates are used in growth media. However it is generally sugars which are held accountable for great growths. Liebmann, Naumann and other workers thus refer to the sugars, especially those from sulphite cellulose wastes and Sphaerotilus is usually one, if not the most important, fungus referred to in their studies.

Besides its high oxygen demand and the mechanical troubles due to its mode of growth and floc formation, it tends to create a septic mud in areas where sludge deposits can be laid down; and there is a strong probability that decomposition of the Sphaerotilus flocs liberates H_2S, with an attendant odor nuisance. There is no previous report of Sphaerotilus as a trouble maker in sulphite cellulose polluted waters in the U. S., although the slime mentioned in the report on the pollution survey of Green Bay, Wisconsin, in 1938-39 may have been Sphaerotilus. No list of references is given for the present report but a partial bibliography may be found in "Studies of Sewage Purification. XIII. The Biology of Sphaerotilus natans Kutzing in Relation to Bulking of Activated Sludge" Reprint 2166 from the Public Health Reports.

The Interstate Technical Advisory Committee, representing Oregon and Washington, maintain and have jurisdiction over the work done at the Columbia River Pollution Laboratory located eight miles up river from Vancouver, Washington. This was headquarters during the present detail. The staff there consisted of Mr. John Lincoln, chemist-in-charge, and Mr. Stanley Jewett,

Biologist. A bacteriologist had been engaged (the previous one having resigned) and arrived on the last day of the writer's stay.

Work was divided three ways. Surveys of the main river were carried out, with attendant surveys of some tributary streams. These were necessarily brief as the Columbia watershed is larger than that of the Ohio, and only a small portion could be visited. A detailed review of the work of the laboratory was undertaken, especially with a view of estimating the value of procedures in vogue and possibly to suggest additional work. Finally, an attempt was made to isolate the troublesome organisms, and to arrive at some conclusions relative to the biological conditions in the river.

Preliminary examinations of the river near the laboratory about eight miles below the sulphite pulp and paper mill of the Crown-Zellerbach Corporation showed large irregular flocs of "sewage fungus" attached to the entire submerged surface of floating logs, stones, ropes--all submerged objects that could be seen. Laboratory examination indicated the flocs were Sphaerotilus natans, and isolation procedures were at once begun, using as a solid medium 1.5% agar made up with filtered river water. It was argued that with heavy growths in the river, such water should prove to be a suitable substrate. This proved to be the case, and in about a week pure cultures were obtained; further tests proved them to be Sphaerotilus natans.

The flocs, however, were mixed. Especially prominent was a filamentous form, showing the same false branching as Sphaerotilus, but smaller in diameter and less dense in floc texture. This also was isolated and is available for use to study its ecology and physiology if necessary. It forms a white colony on agar plates and the colonies are larger than Sphaerotilus colonies. Abundant subsurface growth, a growth rate about twice as rapid as

that of Sphaerotilus, failure of a pure culture to form large flocs under aeration in a liquid medium, and absence of conidium formation are other differences. This new organism appears to be a filamentous bacterium intermediate between the mycoides group and Sphaerotilus but is not one of the Chlamydobacteriales.

Other components of the flocs were diatoms and other algae; protoza, rotifers, insect larvae, worms and much debris. Altoghether the flocs were quite complex in structure, and were tough. None-the-less they were capable of being largely swept out of existence by a flood, since most of the "slime growths" were said to have disappeared during a river rise a short time previous to our visit. Whether they were actually torn loose, or whether the dilution factor cut down their nutrient material and stopped growth is not known. Floating flocs are frequently seen in the river and frames covered with wire gauze catch them. However, fishermens' nets or traps to measure amounts in the water very quickly become fouled because of rapid growth. The conidia or reproductive cells are produced in vast numbers and readily attach themselves to any solid body, as a string. Then, almost overnight, a floc is formed. It must not be lost sight of that we are dealing with a bacterium, and bacterial reproductive rates under what are probably optimum conditions for this particular species--abundant food, O_2, and a suitable temperature.

The species is also widespread in nature--cosmopolitan-, as are so many microscopic organisms. There is no visible Sphaerotilus in the river at Bonneville Dam, yet Bonneville water, enriched with from 10 to 200 p.p.m. of sulphite digester liquor, and allowed to flow through troughs, quickly develops flocs of Sphaerotilus. In other words, the organism is so widespread that wherever suitable conditions are set up, growth of Sphaerotilus

is certain to occur. In the Columbia, the Washington side below Camas (where the Crown-Zellerbach mill is located) is favorable, but the wastes mix slowly, hugging the Washington side. On the Oregon side there is no sulphite liquor and no Sphaerotilus. This is true also at Vancouver, 16 miles below Camas. At St. Helens there is a Kraft paper mill. Here, despite the malodorous condition of the water, its brown color, and the slow dispersal of the wastes because tidal action washes it back and forth, there is no Sphaerotilus. This is a situation where industrial pollution is pronounced, yet conditions are not favorable to Sphaerotilus. The answer probably lies in the absence of sugars in the Kraft mill effluent. There are other paper mills, especially up the Willeamette River, but they were not visited.

A trip was made up the Yakima River, a tributary in Washington. In this valley much fruit is raised, including pears, which are canned. About the time the pear canning is in full swing, a slime fungus, presumably Sphaerotilus, appears in the river and also in municipal sewage disposal plants. None was visible at the time of our visit, except a few scattered flocs in some sewage disposal plants. The extent of the trouble to sewage disposal plants was not ascertained, but where the irrigation ditches lead off from the river, revolving drum screens (Figure 1) have been installed to prevent fish from entering the irrigation ditches and during the pear canning season cleaning the "slime" off these screens is a continuous operation. At the time of our visit the drums had only an occasional bit of debris or water weed on them.

No trip was made to the paper mills on the upper Williamette. It is understood that these mills also contribute wastes which further the development of Sphaerotilus. This valley is heavily populated and the river gets

considerable pollution, the largest amount being along the Portland water-front, where there is a pronounced oxygen sag.

On the whole, the paper mill pollution question does not, so far, materially affect the population as a health problem. No human water supplies are derived, at present, from the streams where such pollution occurs. However there is some question of locating a large army camp on the upper Williamette and using the river as a source of supply. Should this occur, it at once opens up the health problem directly, and it must be determined in advance if possible whether water containing paper mill pollution can be rid of Sphaerotilus and other growths, taste, odor and color with small enough expense and efficiently enough, to make it a potable water.

Nor do the biological effects at present seem to be too harmful. Dilution is great, and if there is no formation of sludge deposits the wastes are soon dissipated to a point where scarcely detectable. It is not known, however, whether sludge banks might be present at some points, and the evolution of gas in Camas slough is certainly evidence of some sludge banks there.

The wastes are not taxic to fish in relatively high concentration. There have been some experiments at Gig Harbor relative to toxic effects on shad eggs, but no opportunity to investigate this work arose. As a partial test of the biological effects, a few plankton examinations of the river were made. Eight of these were Columbia River samples at and below Camas; one was from Bonneville; two were from the Yakima River, and one each from the Washougl, Little White Salmon and Williamette Rivers. The species found, per ml. of raw river water, are shown in table 1. This is a very small amount of data, but it was collected at a favorable time, when the water was low,

and summer conditions prevailed. For this reason it probably illustrates optimum growth condtions of plankton at the points studied. Several points are illustrated therein. (1) It is a typical river plankton. Only one species, Gonyostomum semen, was found, which has not previously been found by us in rivers of the eastern U. S., and this species has been found abundantly in cedar and cypress swamps. In the Columbia, it was found where the brown effluent of the Kraft mill at St. Helens gave the water the same appearance as the swamps. (2) The species list is decidedly smaller than a similar list compiled for Ohio River Basin streams, including the Ohio River proper. There are, of course, streams in the Ohio basin with small plankton populations. Most of the Columbia streams rise in relatively insoluble strata, however, and there is so little pollution that the plankton paucity might be expected. Temperatures for the most part were much lower than in eastern streams also. (3) Total numbers of plankton are small. This is a logical finding. (4) Plankton indicative of pollution is largely absent. Ten or more species of Volvocales were found, but only in small numbers. Furthermore, certain Volvocales favor strawboard waste. (5) A number of species favorable to clean water were relatively abundant. Chrysophyceae, a predominantly clean water group, were largely absent, but their absence does not necessarily have to be accounted for. (6) There is some indication that the pulp mill wastes cause an increase in the plankton content of the river water--that is, they fertilize the river. It would be surprising if this were not the case, considering the large amounts of organic substances as sugars, which are added. The amount of nitrogen added is very small, 0.1% of the digester liquor being proteins, but even this amount is sufficient for growth. If somewhat larger amounts of phosphorus and nitrogen

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were introduced, the waste would be a very rich potential medium; as it is, there is a carbon-nitrogen, or carbon-phosphorus unbalance, probably a limiting factor for the stream organisms.

The survey has brought up a number of interesting problems, some directly connected with the paper pulp industry, others not closely applicable. The major pollution of the Columbia is undoubtedly due to paper mill wastes and any report from the Columbia River Pollution laboratory should include a rather careful study of the biochemical oxygen demand (B.O.D.) of the wastes contributed by each mill. In making this study a mineralized phosphate buffered dilution water should be used. See Sewage Works Journal 13,699 - 1941.

Proper B.O.D. determinations should indicate the origin of the organic wastes in the Columbia River. The difference in the B.O.D. of the river water at the laboratory on the Washington and Oregon sides is significant, these values being about 4 and 1 respectively. It would seem advisable to make a rather careful B.O.D. survey of this section of the river in view of the fact that sewage pollution is practically lacking in the Columbia and that paper mill waste (sulphite) has a B.O.D. many times as great as that of domestic sewage. It is apparent from the lignin determinations that 100 p.p.m. of sulphite liquor might be found in the river on the Washington side and none on the Oregon side below the Crown-Zellerbach mill and these B.O.D. figures might possibly be low. The director of research at the Crown-Zellerbach mill showed a cooperative and friendly interest in the problems involved. In seeking the aid of an industry to solve pollution problems such as this, it seems proper to make the case as strong as absolute facts permit.

It is also desirable to establish criteria of pollution for the Kraft mills, as at St. Helens. There is apparently no Sphaerotilus problem here, but color, odor and debris certainly indicate gross pollution at the point visited. What the criteria of pollution may be we are not prepared to say, but there is some indication that increased plankton growth at this point might be useful. Some comparative studies of species and numbers are certainly indicated for the biologist.

There is also a possible difference from the pollution viewpoint between the magnesium base sulphite process, and the calcium base process. Determinations of chemical and biological differences in river waters below such mills are indicated. Enough determinations should be made of ammonia and total nitrogen and total phosphorus in waters containing paper mill wastes so that the normal relationship of these might be established. The Columbia is a plankton poor river (on the basis of our few determinations) and it is probably low in N, P, and Ca. Sawyer noted that the addition of sewage to paper mill wastes added necessary N and P, but if biological action is to be counted on to help reduce such wastes, there might still be poor biological action in a river poor in calcium. This whole problem of the ratios of these elements necessary to "good" biological action is an intensely interesting one.

It should be determined what part of the sulphite waste promotes the growth of Sphaerotilus (and other organisms, especially floc formers). This involves separating the wastes into fractions and the research division of a paper mill company might well do this. In part it necessitates a review of what sugars are usable by Sphaerotilus et al. The idea that the pentosans are not fermentable might not hold for Sphaerotilus. Bottle experiments

using pure cultures can easily answer this, also the effects of using sulphite wastes enriched with P, N, and Ca. Bottle experiments can also be useful to determine the effects of certain other industrial wastes, as those from pear canning on the growth of Sphaerotilus and troublesome floc forming organisms. And as we have pointed out elsewhere, the floc forming organisms other than Sphaerotilus and Zoogloea should be investigated.

It appears to us that supplementary indices of pollution should be established for all these paper mill wastes. Coliform indices are hardly applicable, but should be known. Biological effects on both plankton and fish or fish eggs might be very helpful in this respect.

There are other problems too, but all are based on the location of proper sampling points, adequate sampling, an appreciation of the differences between the pollutants involved, with the accumulation of adequate chemical, biological and bacteriological data. Some of the above recommendations can undoubtedly be solved by the staff of the Columbia River Pollution Laboratory. It resolves itself into a matter of which of many problems are most pressing.

It must not be inferred from the above considerations that the pulp mill wastes are a nuisance rather than a danger. It has been shown that in the East River and Green Bay, Wisconsin a high B.O.D. and H_2S production resulted from paper mill wastes. There is no doubt that the heavy growths of Sphaerotilus in the Columbia are directly traceable to the Camas mill, and these growths materially affect salmon fishing in the area where they are present. Whether the wastes, tho non-toxic to the salmon, adversely affect the salmon run, should be intensively studied for any factor militating against an industry returning $10,000,000 annually (see the Saturday Evening

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Post for Sept. 13, 1941) is important.

What is more important is that the wastes become a factor changing the river ecology. It is difficult, usually impossible, to foretell the end of one single change in a biologic balance. And if the river is changed slightly by a dam, and slightly by the addition of domestic sewage, and slightly by the admission of paper mill wastes, the effects are cumulative. Presently the salmon are gone; communities wishing to use the river for water must fight expensive tastes, odors, colors and mounting bacterial counts; the river as it was is gone. If there is ever any question of using a river for drinking water, for a food supply (fish), for recreational purposes, it is of the utmost importance that any man-made changes in the river be considered in advance, at great length, and that any water taken from the river be returned in as good or better condition as when it was taken.

(a)

Table 1.

Plankton organisms per ml. of raw river water in the Columbia and certain tributaries in August, 1941.

Stations	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Dates	8-14	8-14	8-17	8-17	8-18	8-18	8-18	8-18	8-19	8-22	8-22	8-22	8-22	8-23	8-23
Organisms - per ml.															
Blue green algae															
Myxophyceae															
Anabaena spiroides			4			3	1	2		3	3	5	4		
Aphanizomenon flos aquae			2	9		4		18		3				1	
Merconiella elegans								3							
Merismopedia tenuissima			2						1		2				
Microcystis incerta										3		3	2		
Oscillatoria sp.	1	12	3								9	4	9		1
Diatoms															
Bacillarieae															
Asterionella formosa				6			14	16		4					
Cyclotella meneghiniana				4			4	4	6	4				2	2
Cymbella sp.							1								
Fragilleria crotonensis										5	5	10	5		
Gyrosigma sp.				1											
Melosira granulata			4		22	32	33	24		14	4	4	8		
" varians													1		
Navicula sp. 40-60 microns			2				4		3	4	2	4	4	1	1
" sp. 15 "									2	4					
Nitzschia sciculoris			4				1			2	3	2	3		
Rhizosolenia eriensis						3									
Stephanodiscus sp.										1					
Synedra actinastroides				3	1	4	8	3		5	1	3	2		
" acus										1	1	1	1	1	1
" ulna			3				1								
Olive green flagellates															
Cryptophyceae															
Chroomonas sp.						5			3	2			2		

Stations	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Dates	8-14	8-14	8-17	8-17	8-18	8-18	8-18	8-18	8-19	8-22	8-22	8-22	8-22	8-23	8-23
				21	16	25	12	33		13	10	9	6	2	2
Cryptomonas erosa										1			1		
" maxima						12			1	6	4	6	3		
Rhodomonas lacustris															
Yellow flagellates															
Chrysophyceae						2				1			2		
Chrysamoeba radians														2	
Chrysococcus amphora					2				4						1
" rufescens										1		1			
Mallomonas sp.															
Dinoflagellates															
Dinophyceae										2					
Gymnodinium gracilis										2	20	19	2		2
Unidentified dinoflagellates		2	2		16	8									
Green flagellates												2			
Euglenophyceae			4												
Euglena intermedia		21								1					
" sp.			1												
Lepocinclis ovum															
Green flagellates										2			2		
Chloromonadida															
Goryostomum semen															
Green flagellates												2			
Chlorophyceae: Volvocales					1	1	6	6		1	2	5			
Carteria large species			2												
" small "			10	33	44	55	84	189	2	10	12	12	18	2	4
Chlamydomonas spp.			1			1						1	1		
Chlorogonium euchlorum			1			1				1					
Collodictyon triciliatum							1						1		
Endorina elegans			1		1										
Gonium pectorale						1							1		
Heteromastix angulosa							1	2		1					
Pandorina morum					2	7	12	6	1			4			
Thoracomonas phacotoides															

Stations	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Dates	8-14	8-14	8-17	8-17	8-18	8-18	8-18	8-18	8-19	8-22	8-22	8-22	8-22	8-23	8-23
Green Algae															
Chlorophyceae															
Actinastrum gracillimum					2	3									
" hantzschii			1				1	2	1	4	1	2	1	1	1
Ankestrodesmus falcatus				6		2	2			2	4				
" " var. mirabilis				30	4	15	39	39	5	22	8	12	12	4	2
Closterium acicularis							1								
Coelastrum microporum			2	12	8	5	12	9	2	12	6	7	7	1	1
" reticulatum										2					
Dictyosphaerium pulchellum			2		6	2	1	1	1	5	6	3	2		1
Francia sp.				6		2				7			6		
Gloeactinium limneticum						1									
Golenkinia radiata					2		2								
Kirchneriella lunaris				3	2					7					
Lagerheimia brevispina										10			2		
Micractinium pusillus					8	3				10	2	2	1		
Mougeotia sp.					3										
Oocystis sp.			4	12	5	8	12	12	2	10	6	8	6	2	2
Pediastrum boryanum							1		1	5	2	1	1		
" duplex										5	4	1	1		
" simplex												1			
" tetras												2	2		
Planktosphaeria gelatinosa										2					
Quadrigula closteroides					1										
Scenedesmus spp.			4	12	16	38	24	63	1	15	12	38	9		1
Schroederia setigera						3						2			
Selenastrum gracile							1								
Staurastrum sp.					1		1								
Tetraedron caudatum										2					
Tetrastrum heteracanthum							6	3							
" staurogeniforme				12			6			7	-	3	2		
Treubaria crassispina						3									

Stations	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Dates	8-14	8-14	8-17	8-17	8-18	8-18	8-18	8-18	8-19	8-22	8-22	8-22	8-22	8-23	8-23
Westella botryoides						3	12						5		
Xanthidium sp.										5					
Green cells, unidentified	18	18	6	2	4	2	6	4	6	10	12	10	8	2	2
Flagellate protozoa															
Mastigophora															
Bicoeca lacustris										18					
Cyathomonas truncata					2										
Desmarella moniliformis					8										
Dinomonas vorax				2								7			
Oicomonas sociabilis						2									
" termo				2						2					
Unidentified flagellates			5							7	3	2	2		1
Amoeboid protozoa															
Sarcodina															
Acanthocystis aculeata										2					
Raphidiophrys pallida					2	10				2			2		
Ciliate protozoa															
Infusoria															
Cyclidium glaucoma			1	2					1	2					1
Oxytricha sp.			1							1					
Strobilidium humile										4		1	1		1
Strombidium gyrans					1										
Tintinnidium fluviatile					1		1	2	1	2	2	2	2	1	1
Urotricha farcta										3			1		
Vorticella sp.			2							1			2		
Rotifera															
Trochelminthes															
Anurea sp.													1		
Rotifera unidentified				1				1		1			1		

Stations

1=Yakima River near Yakima
2=Yakima River near Yakima
3=Willamette River near Portland
4=Columbia River near laboratory
5=Columbia River near Ellsworth
6=Columbia River near Ellsworth
7=Columbia River near Brickyard
8=Columbia River near Camas Slough
9=Washougl near mouth
10=Columbia, Willamette Slough
11=Columbia, Willamette Slough
12=Columbia middle near mouth Lewis
13=Columbia near laboratory
14=Columbia near Bonneville
15=White Salmon near mouth

Figure 1

Revolving drum screens at entrance of irrigation ditches. At pear canning time these become clogged with Sphaerotilus.

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